

A REVIEW ON MICROBIOLOGY AND MOLECULAR BIOLOGY

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ABSTRACT

Forensic microbiology is an old science in a new application. Its introduction to the forensic sciences has been a necessary response to terrorist threats. The benefits that microbiology brings to criminal investigations extend well outside those required for the investigation of terrorism. In return, microbiology has benefited from the enormous advances imposed by the need to develop tools for forensic investigation.

The purpose of this article is to describe how microbiology is applied in the investigation of bioterrorism, highlighting the modern advances in technology, particularly the DNA technologies, which have assisted this discipline as a forensic practice.

Predictive microbiology is based on the premise that the responses of populations of microorganisms to environmental factors are reproducible and that by characterizing foods in terms of those factors, it is possible, from past observations, to predict the responses of those microorganisms in other analogous environments. This knowledge is summarized in mathematical models to enable prediction of the behavior of microbial populations in foods over time. Predictive microbiology is a powerful tool to aid microbial food safety and quality assurance, both in its own right and to complement hazard analysis and critical control points programs, hurdle technology, and quantitative microbial risk assessment.

This article considers the history, philosophy, and impetus of predictive microbiology; principles of mathematical modelling; types of predictive microbiology models; uses, strategies, and resources for 'predictive microbiology'; and assessment of the performance of 'predictive microbiology' models.

Keywords :- Molecular biology , bacteriology , cell biology.

I. INTRODUCTION

- Microbiology is the study of all living organisms that are too small to be visible with the naked eye. This includes bacteria, archaea, viruses, fungi, prions, protozoa and algae, collectively known as 'microbes'.
- microbes play key roles in nutrient cycling, biodegradation/biodeterioration, climate change, food spoilage, the cause and control of disease, and biotechnology.
- Thanks to their versatility, microbes can be put to work in many ways: making life-saving drugs, the manufacture of biofuels, cleaning up pollution, and producing/processing food and drink.

II. METHODOLOGY

- Understanding the principles of microbiology and human cell mechanisms allows pharmacists to discover antimicrobial drugs that would prevent an escalating number of communicable diseases.
- Pharmacists and microbiologists work synergistically to ensure that drug therapies target the opportunistic microbes without harming its human host. Another important role in pharmaceuticals is the use of microbes for the medically important studies.

OSMATIC MICROBIOLOGY

- According to International Microbiology, microbial contamination of cosmetic products is a matter of great importance to the industry and it can become a major cause of both product and economic losses.
- Moreover, the contamination of cosmetics can result in them being converted into products hazardous for consumers.
- The water and nutrients present in cosmetics make them susceptible to microbial growth, although only a few cases of human injury due to contaminated cosmetics have been reported.
- More often, microorganisms are the cause of organoleptic alterations, such as offensive odours, and changes in viscosity and colour.

STERILIZATION

- Sterilization can be defined as any process that effectively kills or eliminates transmissible agents (such as fungi, bacteria, viruses and prions) from a surface, equipment, foods, medications, or biological culture medium.
- In practice sterility is achieved by exposure of the object to be sterilized to chemical or physical agent for a specified time.
- Various agents used as sterilant elevated temperature, ionizing radiation, chemical liquids or gases etc. The success of the process depends upon the choice of the method adopted for sterilization.

III. MODELING AND ANALYSIS

Dry heat Sterilization

- It is well known fact that microorganisms get kill by dry heat due to oxidation effect. Direct flaming direct flaming designer one of the simplest methods of dry heat sterilization in reality the dry heat sterilization is mostly used in microbiology laboratory for the sterilization of the inoculating loops which is Delhi accomplice by heating the loop wire to a red glow and this is 100% effective in actual practice likewise the same principle is even extended to the process of incineration to Sterilize as well as dispose of heavy contaminated paper bags cups and dressings.

Hot air sterilization

- It may be regarded as another kind of dry heat sterilization in this particular process of various items need to be sterilized are done in electric oven prepared with stainless steel chamber inside and Dolly maintain at 170 degree Celsius for duration of approximately 2 hours to ensure complete sterilization.
- It has been adequately observed that the longer the period + higher temperature are needed proposedly due to the fact that the heat in water is more rapidly passed on to a cool body in comparison is the heat in air.
- Disinfectants are substances that are applied to non-living objects to destroy microorganisms that are living on the objects. Disinfectants are substances that are applied to non-living objects to destroy microorganisms that are living on the objects.

AOAC Method:-

- The AOAC dilution method is a standard currently being employed for the evaluation of disinfectant methodology three strains of microorganisms are usually employed in the AOAC method such as

salmonella cholerae Suis Staphylococcus aureus and Pseudomonas aeruginosa the various steps in all are the follows:

- To carry out use dilution, take the metal carrier rings are dipped deep into the standard culture of the test organism adequately grown in a liquid media removed carefully write at 37 degrees Celsius for a short duration.

Filter Paper Method

- The filter paper method is commonly used in the effects evaluation of a chemical agent as a disinfectant in teaching practice in Laboratories a small disk of filter paper preferably what man grade is duly soaked in a solution of a chemical agent and placed expectedly on the surface of other plate which has been previously encultured and incubated so you live with a pure test organism the effect to be exhibited by a clear zone as the zone of any vision designating precisely the initiation of growth just around the disk.

Disinfectants Variant:-

- 1) Alcohol
- 2) Aldehyde
- 3) Halogen
- 4) Oxidizing agent
- 5) Surface Active agent

DNA

- ✓ DNA is a polymer composed of two polynucleotide chains that coil around each other a form of double helix the polymer carries genetic instruction for the development function in growth and reproduction of all non-organisms and viruses DNA and Ribonucleic acid nucleic acid alongside proteins lipids and complex carbohydrates polysaccharide nucleic acid one of the four major types of micro molecules that are essential for all known forms of life.
- ✓ The two DNA strands are known as polynucleotides as they are composed of simpler monomeric unit called nucleotides a nucleotide is composed of one of four nitrogen containing nucleotide bases Cytosine Guanine Adenine Thymine a sugar called as the deoxyribose phosphate group. then nucleotides are joined to one another in a chain by covalent bonds are also known as phosphodiester linkage.
- ✓ The nitrogenous base of the two separate polynucleotide strands are bound together according to base pairing rules A with T and C with G hydrogen

bonds to make double standard DNA the complement nitrogenous bases are divided into two groups the single ring pyrimidines and the double rings purines the pyrimidine is the thymine and cytosine and adenine and guanine.

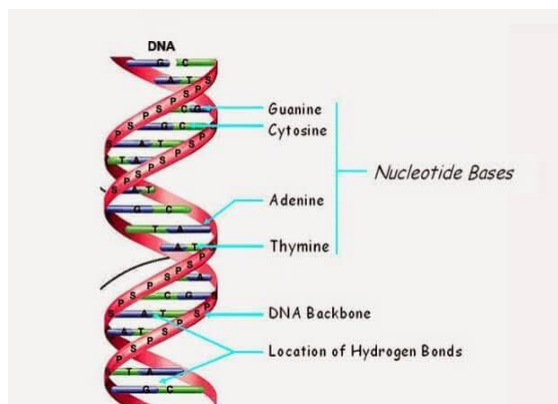


Figure 1:- Structure of DNA

RNA

- Ribonucleic acid RNA is a polymeric molecule that is essential for most biological functions either by performing the function itself non coding rna forming a template for the production of proteins messenger RNA.
- RNA and deoxyribonucleic acid and nucleic acid . the nucleic acid constitutes one of the four major macro molecules essential for all known forms of life RNA is assembled as a chain of nucleotide cellular organism messengers RNA. to convey genetic information using the nitrogenous bases of the letter Guanine Uracil Adenine Cytosine that directs synthesis of specific proteins many viruses in court their genetic information using RNA genome.

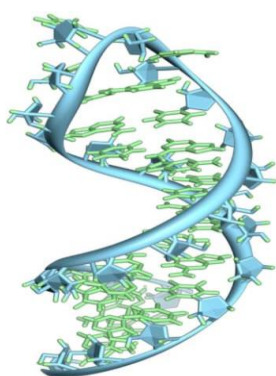


Figure2:Structure of RNA

Proteins

- Proteins are the end products of the decoding process that starts with the information in cellular DNA. as work horses of the cell protein compose

structural and motor element in the cell and they serve as the catalyst for Virtually every biochemical reaction that occurs in living things incredible array of function derives from a strolling simple code that specifies a hugely diversity of structure.

- In fact, itching in cellular DNA conduct the code for unique protein structure not only are these proteins assemble with different amino acid 6 phase but they also are 1 Together by different bonds and folded into variety of three-dimensional structure the folded shape or confirmation depends directly on the linear Ammonia acid sequence of the protein.
- The building blocks of protest around us in which are small organic molecules that consist of Alpha Central carbonate am into an amino group A carboxyl group hydrogen atom and horrible component call side chain within a protein multiple hour acids are linked Together by peptide Wars there by forming a long chain bonds are formed by chemical reaction that extract water molecules acid joints the amino group of one acid to the carboxyl group of neighbouring our linear equation a protest president of the proteins.

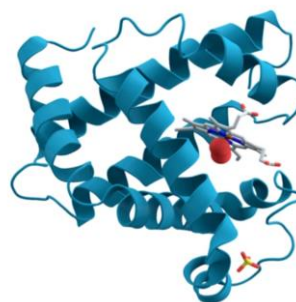


Figure3:-Structure of Protein

Insulin

- The role of insulin in the body is to allowed glucose in the blood to enter the sales providing them with the energy to function a lack of effect to insulin play the key role in the development of diabetes insulin is a type of hormone hormones are chemical messengers that instruction sales or tissue to act in a certain way that supports a particular function in the body insulin essential for staying alive insulin is a chemical messenger that all of sales to abs of glucose a sugar from the blood the pancreas is an organ behind the stomach that is the main source of insulin in the body cluster of sales in the pancreas called isolate produce the hormone and determine the amount base on blood glucose levels in the body the higher the level of glucose the more insulin goes into production to balance sugar levels in the body insulin also assist in Breaking Down fats or proteins

for energy the delicate balance of insulin regulated blood sugar and many process in the body if insulin levels are to lower high excessively high or low blood Sugars start to cause symptoms.

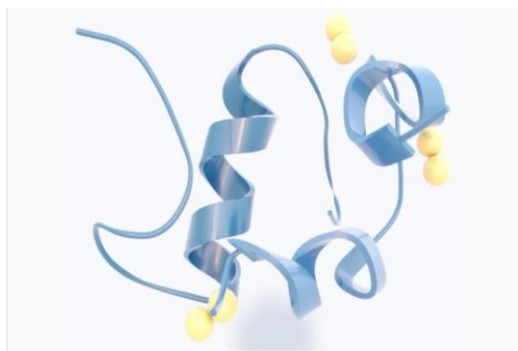


Figure 4:- Structure of Insulin

1. Entry

- 1) Enter the quality control department through the staircase
- 2) remove the street footwear and keep them in the designated place.
- 3) cross over the bench and were dedicated QC slippers
- 4) open the garment cubical pick and wear dedicated by white apron
- 5) open the entry door and reach the microbiology selection through QC corridor
- 6) press interlocking but open the airlock door
- 7) open the entry door and enter into the air lock of microbiology section remove the dedicated QC slippers and keep them in designated place.
- 8) cross over the bench and wear the dedicated microbiology lab slippers
- 9) press the interlocking button and open the door and enter into microbiology lab general coriander.

2.Exit:-

- 1) Press interlocking button to open the air lock door.
- 2) open the exit door of main entry airlock and enter into the air lock keep the dedicated microbiology lab slippers in the designated place in the case of shoes covers remove the covers and put them into the dustbin
- 3) cross over the bench and wear dedicated QC slippers .
- 4) press the interlocking button and open the door leading to exit from the QC
- 5) cross over the bench and wear street footwear

6) open the exit door and reach the stair case to department for the QC department

7) in the case of dedicated factory dress in the change room and wear the street garments.



Figure 5:-Microbiology Laboratory

SOME LABORATORY INSTRUMENTS ARE USED IN LABORATORY

- 1) Petri Dish
- 2) Spatula
- 3) Wire Brush
- 4) Hot plate
- 5) Pipette
- 6) Agar Slant
- 7) Measuring Jar
- 8) Measuring Cylinder
- 9) Funnel
- 10) Incubator
- 11) Micro Pipette
- 12) Lab Coat
- 13) Burner
- 14) Forceps
- 15) Tripod

IV. RESULT AND DISCUSSION

List of media:

- Differential media
- The differential media usually request to the incorporation of certain specific Chemicals into medium that may even showily give rise to diagnostically useful growth or Apparent change in the membrane after the proper incubation a few typical examples are discuss under following

➤

1) Eosin Methylene Blue Agar (EMB Agar)

- The EMB Agar media is employed exclusively to differentiate between the lactose fermentation and the non-lactose fermenters.

- inside the EMB Agar media essentially comprise of the Lactose Salts and two dye eosin and methylene blue. from the observation the interfere following.

- 1) E coli will produce either dark Colony or one of that has media sheen and
- 2) S typhi shall appear as an absolute colourless Colony.

Enrichment Media

- It has been Amply demonstrated and established the critical and judicious incorporation of blood or extract to the particular tryptic soy Agar or broth shall enormously argument the desired growth of a large number of most fastidious microbes.
- In actual practice however largely employed to isolate Primarily the microorganisms from host of biological such as cerebrospinal fluid pleural fluid wound abscesses and sputum.

1) Blood Agar

- The critical addition citrated blood to the prevailing tryptic soya gardeners it is the variable haemolysis that in turn all of the precious differentiation of certain species of microorganisms it is how your pertinent to state here that one May observe this thing haemolytic patterns of the Other a few searches typical variations are stated under.



Figure 6: Blood Agar

a) Alpha Haemolysis: -

- It may be observed due to the formation of greenish to brownish around the colony example streptococcus gardenia and streptococcus pneumoniae.

b) Beta Haemolysis: -

- It represents the virtual complete haemolysis of blood cells there by giving rise to a distinct clearing effect around growth in the colony example staphylococcus aureus and staphylococcus pyrogens.

c) Nonhemolytic:

- Pattern in this particular instance participatively no change ours in the medium example Staphylococcus epidermidis and Staphylococcus saprophyticus.



Figure 7: Different types of culture media

TESTS

BACTERIAL ENDOTOXIN TEST

Endotoxin:

- Endotoxin alkyl polysaccharide is a pyrogenic substance that is found in the cell wall
- Gram-negative bacteria.
- Pyrogenic substance (or pyrogen) can induce fever when injected into the blood or cerebrospinal fluid
- It is associated with injectable products
- Sterile production procedures are needed
- Sterilization does not remove the endotoxin

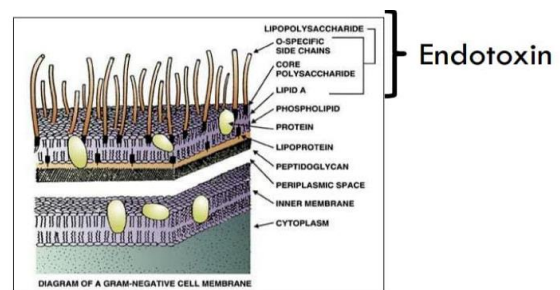


Figure8: Endotoxin Test

Bacteria endotoxin test

- Bacterial endotoxin test (aka LAL test): -
- To detect or quantify endotoxin of gram-negative bacterial origin using amoebocyte lysate from horseshoe crab

Types of Lal test:

Methods: -

- ✓ Gel clot
- ✓ Gel clot (Limit test)
- ✓ Gel clot (Semi-quantitative test)
- ✓ Photometric
- ✓ Chromogenic (Kinetic)
- ✓ Turbidimetric (Kinetic)

ELISA TEST INFORMATION

- It is commonly used analytical biochemistry assay, first described by Eva Engvall and Peter Pearlman in 1971. The assay uses a solid-phase type of enzyme immunoassay (EIA) to detect the presence of a ligand (commonly a protein) in a liquid sample using antibodies directed against the protein to be measured. ELISA has been used as a diagnostic tool in medicine, plant pathology, and biotechnology, as well as a quality control check in various industries.
- In the simplest form of an ELISA, antigens from the sample to be tested are attached to a surface. Then, a matching antibody is applied over the surface so it can bind the antigen. This antibody is linked to an enzyme and then any unbound antibodies are removed. In the final step, a substance containing the enzyme's substrate is added.
- A buffered solution of the antigen to be tested for is added to each well (usually 96-well plates) of a microtiter plate, where it is given time to adhere to the plastic through charge interactions.
- A solution of nonreacting protein, such as bovine serum albumin or casein, is added to each well in order to cover any plastic surface in the well which remains uncoated by the antigen.
- The primary antibody with an attached (conjugated) enzyme is added, which binds specifically to the test antigen coating the well.
- A substrate for this enzyme is then added. Often, this substrate changes colour upon reaction with the enzyme.
- The higher the concentration of the primary antibody present in the serum, the stronger the colour change. Often, a spectrometer is used to give quantitative values for colour strength.

Enzyme Immobilization:-

- Immobilization of enzymes (or cells) refers to the technique of confining/anchoring the enzymes (or cells) in or on an inert support for their stability and functional reuse. By employing this technique, enzymes are made more efficient and cost-effective for their industrial use. Some workers regard immobilization as a goose with a golden egg in enzyme technology.
- Immobilized enzymes retain their structural conformation necessary for catalysis.

Methods of Immobilization:

1) Adsorption:

- Adsorption involves the physical binding of enzymes (or cells) on the surface of an inert support.
- The support materials may be inorganic (e.g. alumina, silica gel, calcium phosphate gel, glass) or organic (starch, carboxymethyl cellulose, DEAE-cellulose, DEAE-Sephadex).
- Adsorption of enzyme molecules (on the inert support) involves weak forces such as van der Waals forces and hydrogen bonds. Therefore, the adsorbed enzymes can be easily removed by minor changes in pH, ionic strength or temperature.

2) Entrapment:

- Enzymes can be immobilized by physical entrapment inside a polymer or gel matrix.
- The size of the matrix pores is such that the enzyme is retained while the substrate and product molecules pass through
- In this technique, commonly referred to as lattice entrapment, the enzyme (or cell) is not subjected to strong binding forces and structural distortions.
- Some deactivation may however, occur during immobilization process due to changes in pH or temperature or addition of solvents. The matrices used for entrapping of enzymes include polyacrylamide gel, collagen, gelatin, starch, cellulose, silicone and rubber. Enzymes can be entrapped by several ways.

3) Microencapsulation:

- Microencapsulation is a type of entrapment. It refers to the process of spherical particle formation wherein a liquid or suspension is enclosed in a semipermeable membrane. The membrane may be polymeric, lipoidal, lipoprotein-based or non-ionic in nature.

There are three distinct ways of microencapsulation.

1. Building of special membrane reactors.
 2. Formation of emulsions.
 3. Stabilization of emulsions to form microcapsules.
- Microencapsulation is recently being used for immobilization of enzymes and mammalian cells. For instance, pancreatic cells grown in cultures can be immobilized by microencapsulation. Hybridoma cells have also been immobilized successfully by this technique.

4) Covalent Binding:

- Immobilization of the enzymes can be achieved by creation of covalent bonds between the chemical groups of enzymes and the chemical groups of the support.
- This technique is widely used. However, covalent binding is often associated with loss of some enzyme activity.
- The inert support usually requires pretreatment (to form pre-activated support) before it binds to enzyme. The following are the common methods of covalent binding.

5) Cross linking:

- The absence of a solid support is a characteristic feature of immobilization of enzymes by cross-linking.
- These reagents in fact react with the enzyme molecules and create bridges which form the backbone to hold enzyme molecules.
- There are several reagents in use for cross-linking.
- These include glutaraldehyde, diazo benzidine, hexamethylene diisocyanate and toluene.

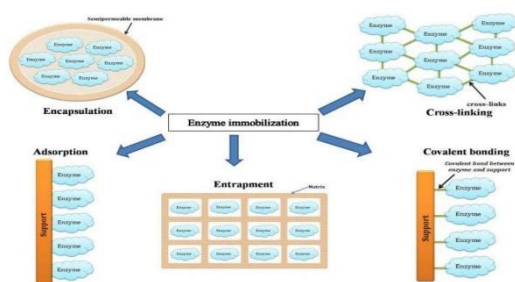


Figure 8: Enzyme immobilization

V. Conclusion:

Microbiology plays a crucial role in terms of preventing some kind of infection and bacteria from the living being. Thus, it can be concluded that microbiology plays a crucial role in terms of preventing some kind of infection and bacteria from the living being and that can be helpful for the future biological perspective. Microbiology plays a crucial role in medical microbiology and why mental microbiology and the Food Industry can be significantly helpful for preventing bacteria and viruses from foods and human beings. It helps to identify the treating diseases of the human body and agricultural perspective that can be applied to find the specific cells in a Complex biological system. There is a high scope in the field of microbiology which can be helpful in the induction of modern life. Molecular microbiology deals with molecular mechanisms and physiological processes of microbes and their utilisation in production of

biotechnology products and medicines such as vaccines, antibodies.

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